Influence of the Chemical Environment on Metolachlor Conformations

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Metolachlor exists in multiple, different stable conformations in solution. Assignment of the NMR frequencies to chemical structure is a prerequisite to understanding the behavior of individual conformations. ^{1}H NMR experiments of metolachlor in different chemical environments identified the labile sites of metolachlor and environments that influence conformational/configurational changes. Within very specific chemical environments, metolachlor atropisomers aS,12S (aR,12R) and aR,12S (aS,12R) freely interchange, and consequently, the multiple conformations also interchange. The changes in chemical environments, which most alter the conformations and molecular dynamics of metolachlor, identify the most critical components affecting its environmental fate. These results enable a structural interpretation of conformational changes that can influence the environmental fate of metolachlor.

INTRODUCTION

The environmental fate of pesticides and other organic molecules depend on their interactions with the local chemical environment at the molecular level. Most organic molecules are conformationally flexible, with the active conformations being only a few of the accessible conformations in solution. The conformations of organic molecules can be influenced by their chemical environment, which will affect environmental fate (Schmidt et al., 1995, 1997). The bioavailabilty of an organic pollutant is a function of its sorption to soil constituents. Understanding the molecular features of pollutant sorption is important in predicting availability and degradation mechanisms and rates.

Nuclear magnetic resonance (NMR) spectroscopy is widely used in routine structure elucidation of natural organic matter (Wershaw et al., 1985; Noyes and Leenheer, 1989; Olk et al., 1995; Preston, 1996) and other organic compounds (Mesilaakso, 1997; Morton et al., 1997; Jackson and Line, 1997). Its application in the field of environmental sciences however is relatively untapped. Recent work has proved NMR to be invaluable at identifying conformations and metabolites (Schmidt et al., 1995; Morton et al., 1997), interactions (Hatcher et al., 1993; Nanny et al., 1997; Kim et al., 1997), reactivity, and heterogeneous catalytic processes (Ansermet et al., 1990; Carrado et al., 1990) of organic pollutants. Yet, before NMR can be applied to field conditions, in-depth studies must be conducted in simple chemical environments to provide background information that can be used to interpret interactions in more complex environments.

Metolachlor [2-chloro-N-(2-ethyl-6-methylphenyl)-N-(2-methoxy-1-methylethyl) acetamide] is a preemer-

gence herbicide used to control most annual grasses and broad leaf weeds in beans, corn, cotton, potatoes, etc. (Montgomery, 1993). It is one of the most widely used pesticides in the U.S. agricultural crop production (Aspelin, 1994). In general, the chloroacetamides inhibit the biosynthesis of fatty acids, lipids, proteins, isoprenoids, flavonoids, and terpensids (Ahrens, 1994). Recent surveys have detected metolachlor in surface waters (Godfrey et al., 1995) and groundwaters (Koterba et al., 1993; Ritter et al., 1994). A key process controlling the fate of metolachlor is its interactions with soil constituents such as the organic matter and clay surfaces. These interactions will influence its dissipation from soil. Microbial degradation is the major pathway through which metolachlor is dissipated from soil (Montgomery, 1993). Biodegradation has shown promising results in the remediation of metolachlor-contaminated soils (Dzantor et al., 1993). Hence, it is important to understand the bioavailabilty of metolachlor in soil.

Metolachlor sorption to pure surfaces and soil constituents has been the topic of numerous previous studies (Pusino et al., 1992; Torrents and Jayasundera, 1997; Torrents et al., 1997). These studies focused on macroscopic scale information where sorption was described through sorption isotherms with no information on microscopic level studies that describe specific surface interactions and the different configurations and conformations.
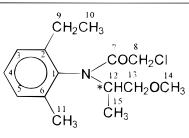
Metolachlor exists as a racemic mixture of two sets of enantiomers (aS,12S/aR,12R and aR,12S/aS,12R) that are due to the chirality at C-12 in the alkyl moiety and the hindered rotation about the Ar–N bond and the asymmetrically substituted aromatic ring (Table 1). A diastereomeric relationship exists between the atropisomers aS,12S and aR,12S (set 1) and aR,12R and aS,-12R (set 2). Set 1 is the mirror image of set 2. The energy barrier for rotation around the Ar–N bond (activation energy = 154 kJ mol^{-1}) (Moser et al., 1982)

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Table 1. Physical and Chemical Properties of Metolachlor (Montgomery, 1993)

Pesticide	Structure	Properties	
Metolachlor		Mol. weight	283.8
		Physical state	liquid
		Boiling point (^0C)	100
		Solubility (mgL^{-1})	530
		log K_{ow}	2.93
		Mol. surface area[a] (A^0)^2	327.9

*Chiral Center

[a] The surface area was calculated using Hyper-Chem using the van der Waals interactions method.

is high; thus, metolachlor atropisomers a*S*,12*S* (a*R*,12*R*) and a*R*,12*S* (a*S*,12*R*) are distinguishable in NMR spectra. Interchange between the metolachlor diastereomers (or rotation about the Ar—N bond) results in a configurational change and is only possible by either overcoming the energy barrier to rotation or decreasing the activation energy.

The chirality of metolachlor can influence its properties. Moser et al. (1982) reported that the *S*-configuration of C-12 has the highest herbicidal activity while the *R*-configuration showed some fungicidal activity. Metolachlor has been shown to have stereo and enantioselective degradation properties in a sewage sludge and soil where the a*R*-configuration was preferred over the a*S*-configuration in a set of enantiomers (Muller and Buser, 1995).

Metolachlor and most pesticides possess functional groups that are sensitive to different chemical environments. Similar to natural microenvironments, different solvents can interact with the functional groups through hydrogen bonding, polar effects, anisotropic interactions, etc. (McClure, 1996).

In this paper, we describe the configurations and different solution conformations of metolachlor, which have not been previously characterized, using NMR and molecular mechanics. We have also studied the influence of solvents and mixtures of solvents to understand the most labile sites, the conditions under which configurational changes can occur, and the resulting final conformations of metolachlor in different chemical environments.

MATERIALS AND METHODS

Chemicals. Analytical grade metolachlor was obtained gratis from Ciba-Geigy Corporation with a purity of 99.0% and was used without further purification. The physical and chemical properties of metolachlor are listed in Table 1. The deuterated (D) solvents chloroform (CDCl$_3$), deuterium oxide (D$_2$O), and dimethyl sulfoxide (DMSO-d_6) were purchased from Isotec, Inc., Miamisburg, OH (each 99+% D). Benzene (Bz-d_6) (100% D) was purchased from Aldrich, Milwaukee, WI. Solvent mixtures were made on a volume basis. All solvents were used as purchased.

Molecular Mechanics. The different stable conformations of metolachlor that are low in potential energy and corresponding to a global minimum were determined using molecular mechanics in the MM+ force fields in HyperChem from Hypercube, Inc. Waterloo, Ontario, Canada. After selecting an initial structure, small systematic variations were applied to the torsion angles. Geometrical optimization was used to find a local minimum on the potential energy surface of each new structure. The conjugate gradient (Polak—Ribiere) method was used as the optimizer, and a RMS gradient of 0.01 was used for considering an optimization to be converged. Geometry and energy comparisons were made with previously accepted conformations to avoid duplication. Torsion angle variations less than 5° and energies within 0.05 kcal/mol were considered

as duplicate structures. Each torsion angle was varied from ± 10 to 180°.

NMR Spectroscopy. Solution NMR spectra were recorded using a Bruker QE Plus spectrometer at 300 MHz for ^{1}H and 75 MHz for ^{13}C. Proton spectra were acquired with a spectral width of 2400 Hz and 4K data points. The carbon spectral width was 24000 Hz with 16K data points that was zero-filled to 32K before Fourier transformation. Two-dimensional homonuclear correlation spectroscopy (COSY) and heteronuclear correlation spectroscopy (HETCOR) confirmed the intramolecular assignments of the chemical shifts for the conformers. One-dimensional nuclear Overhauser enhancement (NOE) difference spectra (NOEDS) experiments and 2D nuclear Overhauser enhancement spectroscopy (NOESY) were conducted to obtain specific conformations. The chemical shifts were referenced against the residual proton signal of the deuterated solvent. The NMR spectra were collected at 298 K and were controlled to ±0.5 of set temperature.

RESULTS AND DISCUSSION

Metolachlor Conformations from Molecular Modeling. The minimum energy conformations of metolachlor determined, in the gas phase, using MM+ force fields in molecular mechanics for both the *S*- and *R*-chiral molecules are shown in Figure 1. Molecular mechanics confirmed a hindered rotation about the Ar—N bond, resulting in increased steric hindrances when the aromatic plane deviated from ∼90° to the amide plane. Moser et al. (1982) determined this energy barrier to rotation as 36.8 kcal mol^{-1} (154 kJ mol^{-1}).

Our results also confirmed that two stable energy-minimized conformations were present when the Ar—N—C=O torsion angle was ∼0° and ∼180°, and deviation of the torsion angle from these angles caused an increase in energy and hence an energy barrier to rotation about the C—N amide bond. The partial double-bond character of the amide bond hinders free rotation around this bond. The carbonyl in the trans position to the aromatic ring was the lower energy conformation (6.94 vs 19.0 kcal mol^{-1} in the *R*-chiral conformer). Using the Boltzman equation (Harris, 1986), it was confirmed that the higher energy population was almost zero and the carbonyl was almost always in the trans position. This is consistent with previously reported observations for substituted amides where the trans position of the C=O to the aromatic ring is preferred and the energy barrier to rotation is 16—21 kcal mol^{-1} (66—88 kJ mol^{-1}) (Brown et al., 1968; Eliel and Wilen, 1994).

Three stable conformations of the isopropyl methyl ether substituent were obtained for each metolachlor stereoisomer (i.e., a*S*,12*S*; a*R*,12*S*; a*S*,12*R*; a*R*,12*R*), which resulted mainly in the three different conformations of each isomer (designated conformations a—c in Figure 1). The molecular conformations of the isomers a*R*,12*R* and a*S*,12*R* are the exact mirror images of a*S*,12*S* and a*R*,12*S*, respectively. The minimum energy conformations in the gas phase of the enantiomers a*S*,12*S*/a*R*,12*R* and a*R*,12*S*/a*S*,12*R* were a and b, respectively.

Metolachlor Conformer and Configuration Structures in Solution. ^{1}H and ^{13}C NMR data for metolachlor were obtained in DMSO-d_6, CDCl$_3$, and Bz-d_6 (Figure 2, Table 2). The proton signals were unambiguously assigned, and the coupling of neighboring protons described by COSY experiments. Two signals of different intensity were observed for the Ar—**CH$_3$** (H-11) protons in all three solvents (Figure 2). The upfield signal was more intense with the intensity ratio be-

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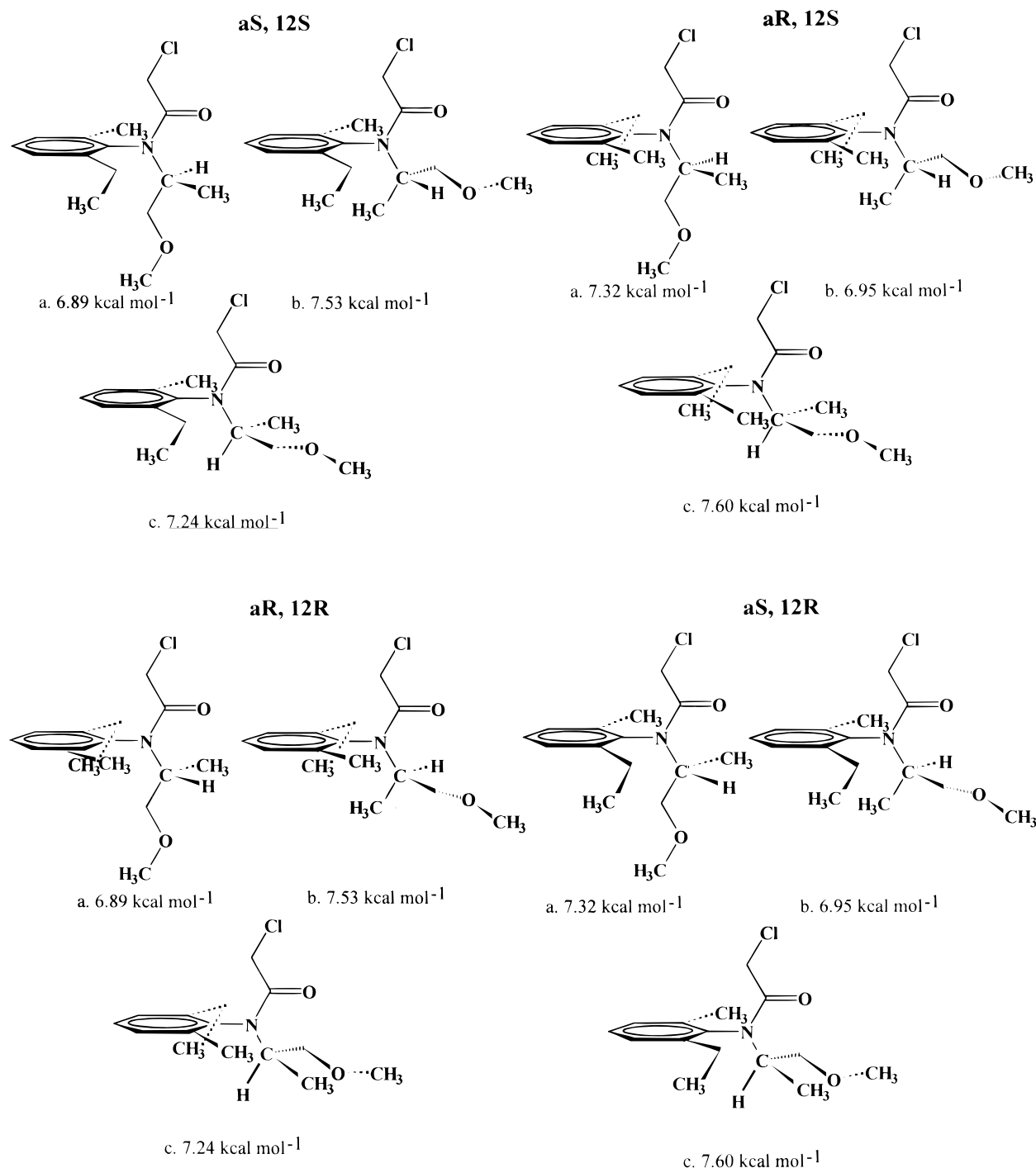*J. Agric. Food Chem.,* Vol. 47, No. 10, 1999 **4437**

Figure 1. Energy-minimized conformations of metolachlor isomers determined from molecular mechanics using HyperChem.

tween the two signals being 31:69 in all three solvents. Due to the asymmetric C-12 of the alkyl moiety and the rotational barrier at the Ar−N bond, the two Ar−**CH₃** signals indicate the presence of two stable configurations. The high and low intensity signals have been identified as a*S*,12*S*/a*R*,12*R* and a*R*,12*S*/a*S*,12*R*, respectively (Moser et al., 1982; H. Sauter, personal communication).

Two-dimensional NOESY and 1D NOE experiments were used to identify nuclei that were close in space when metolachlor was in Bz-*d₆*. NOE effects are generally observed when the spatial distance is <5 Å through dipolar interactions between nuclei. The cross-peaks in the NOESY spectra and NOE experiments (data not shown) show closeness in intramolecular spatial dis-

tance between the H-11 (Ar−**CH₃**) protons of a*S*,12*S*/ a*R*,12*R* and both the −N−CH−**CH₃** (H-15) and −N− **CH**−CH₃ (H-12) protons, strongly suggesting that the c conformation is the preferred conformation by these isomers.

Positive NOE effects were observed at the H-11 signal of a*R*,12*S*/a*S*,12*R* when the H-12 proton was irradiated. Therefore, b and c conformations would be favored for a*R*,12*S*/a*S*,12*R*. However, when −CH−**CH₂**−O− (H-13) protons were irradiated, a positive NOE was observed on the Ar−CH₂−**CH₃** (H-10) protons, and no NOE was observed on the H-11 protons of both sets of enantiomers. Only in the b conformation of a*R*,12*S*/a*S*,12*R* would these NOE effects be possible. These data also further support c as the preferred conformation of
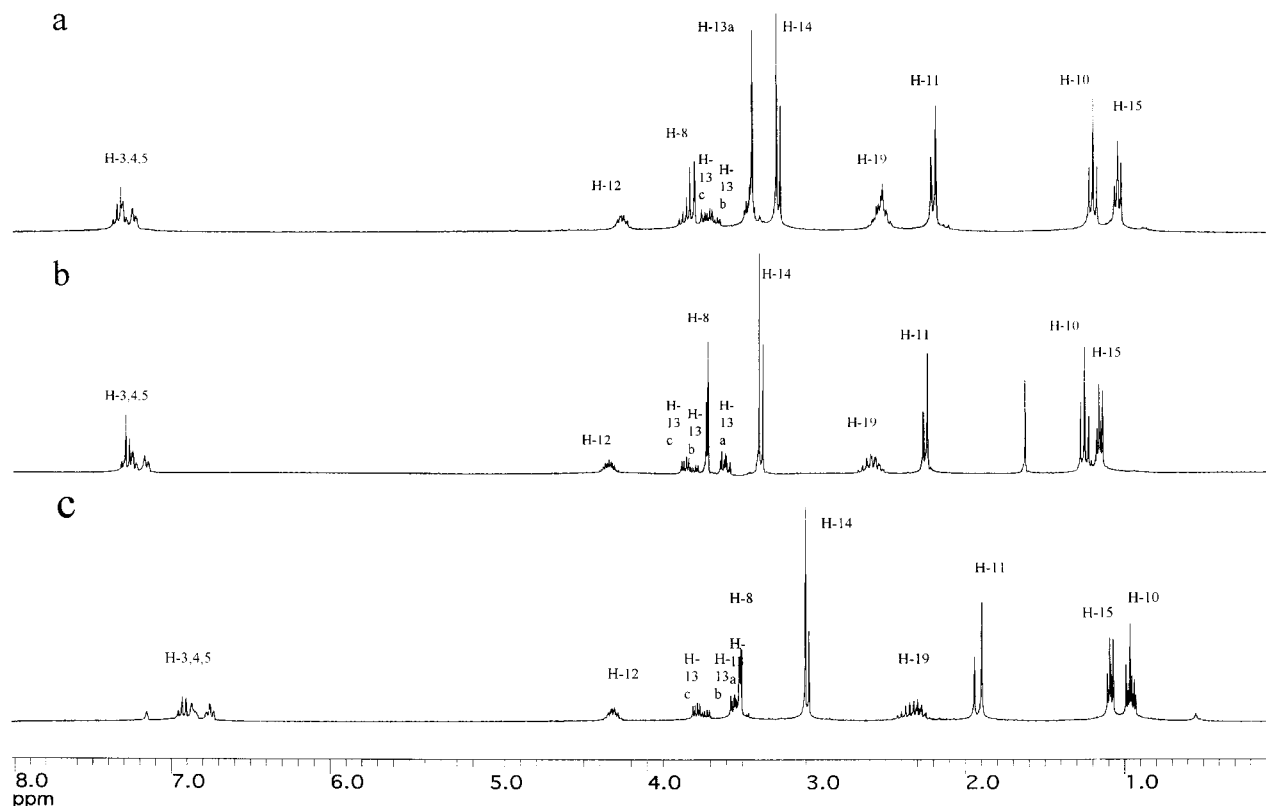
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Figure 2. Proton spectra of metolachlor conformers: (a) in DMSO-d_6; (b) in CDCl$_3$; (c) in Bz-d_6.

Table 2. Proton and Carbon Chemical Shift Assignments of Metolachlor Rotamers and Conformers

chemical moiety[a]	spectra in DMSO-d_6 ^{1}H NMR δ (ppm)	spectra in CDCl$_3$ ^{1}H NMR δ (ppm)	spectra in benzene-d_6 (100%)	
			^{1}H NMR δ (ppm)	^{13}C NMR δ (ppm)
C,H-15 N−CH−**CH$_3$**	1.00 (d)	1.11 (d)	1.05 (d)	15.5
	1.05 (d)	1.15 (d)	1.09 (d)	15.7
C,H-10 Ar−CH$_2$−**CH$_3$**	1.19 (t)	1.23 (t)	0.91 (t)	13.9
			0.94 (t)	14.1
C,H-11 Ar−**CH$_3$**	2.16 (s)	2.21 (s)	1.88 (s)	18.8
	2.19 (s)	2.25 (s)	1.93 (s)	18.9
C,H-9 Ar−**CH$_2$**−CH$_3$	2.52 (m)	2.57 (m)	2.29 (m)	23.9
			2.36 (m)	24.1
C,H-14 −O−**CH$_3$**	3.11 (s)	3.24 (s)	2.96 (s)	55.9
	3.15 (s)	3.28 (s)	3.00 (s)	55.6
C,H-13 −CH−**CH$_2$**−O−	3.33 (m)	3.46 (m)	3.44 (m)	74.9
	3.56 (m)	3.70 (m)	3.60 (m)	
	3.59 (m)	3.76 (m)	3.66 (m)	
C,H-8 CO−**CH$_2$**−Cl	3.67 (s)	3.57 (s)	3.39 (s)	42.7
	3.70 (s)	3.59 (s)	3.41 (s)	
	3.75 (s)		3.42 (s)	
	3.78 (s)		3.42 (s)	
C,H-12 −N−**CH**−CH$_3$	4.13 (m)	4.19 (m)	4.22 (m)	58.3
C-7 −**CO**−CH$_2$−Cl				166.1
				166.2
C,H-3, 5 Ar−**CH** (o) (p)	7.21 (d), 7.26 (d)	7.11 (d), 7.19 (d)	6.87 (d), 6.73 (d)	128.8, 126.7
C,H-4 Ar−**CH** (o) (p)	7.29 (t)	7.25 (t)	6.92 (t)	128.5
C-2 Ar−**C**				137.2, 137.3
C-6				142.8, 142.9
C-1				138.2
residual solvent	2.50 (m)	7.25 (s)	7.15 (s)	128 (t)
	3.31 (s)	1.56 (s)	0.50 (s)	

^{a} Numbering scheme is given in Table 1.

aS,12S/aR,12R. Since NOE data are based on an average interproton distance, the observed NOE effects reflects a population-weighted average closeness between atoms in space of each of the conformations (Bruch and Dybowski, 1996). Therefore c and b are the mostly favored conformations by the enantiomers aS,-12S/aR,12R and aR,12S/aS,12R, respectively, in a solvent medium. This is further confirmed in the following

sections when we describe some of the proton frequencies in metolachlor ^{1}H NMR spectra.

Terminal CH$_3$. The terminal CH$_3$ on the ethyl group attached to the aromatic ring (H-10) shows a single triplet in CDCl$_3$ and DMSO-d_6. This indicates that the two signals for the enantiomers aS,12S/aR,12R and aR,-12S/aS,12R fortuitously occur at essentially the same frequency. However, two sets of triplets were observed

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for H-10 protons in Bz-d_6 (Figure 2; Table 2). This effect can be explained via two mechanisms. The two sets correspond to (1) two minimum energy conformers of the CH_3, obtained above and below the aromatic ring for each metolachlor conformation from molecular mechanics (not shown), and/or (2) two preferred conformations of the two sets of enantiomers (i.e., c for a*S*,12*S*/a*R*,12*R* and b for a*R*,12*S*/a*S*,12*R*). The NOE effects that were observed only in the high-intensity signal of H-10 when irradiated at the H-12 proton and that on both H-10 signals when irradiated at H-13 protons support the latter.

The similar intensity ratios between the two sets of triplets for H-10 and those of the two signals of H-11 protons that represent the two sets of enantiomers also support the second mechanism. The NOESY spectra cross-peaks indicate closeness in intramolecular spatial distance between the lower intensity H-10 proton frequency and $-N-CH-CH_3$ (H-15) protons (described later) of a*R*,12*S*/a*S*,12*R*. This supports mechanism 2 as well as the conformation b for a*R*,12*S*/a*S*,12*R*.

The two frequencies of unequal intensities for the CH_2Cl (H-8) protons in CDCl$_3$ can be explained by the Ar$-CH_3$ rotamer configurations. In DMSO and Bz-d_6, the H-8 protons appear as four frequencies of unequal intensities. Anisotropic interactions between the π-bonds, i.e., S=O in DMSO and C=O in metolachlor and the aromatic rings of both benzene and metolachlor, can induce a cone of shielding/deshielding. These anisotropic interactions may resolve the two hidden frequencies of the H-8 protons in CDCl$_3$ that could correspond to the two H-10 proton conformers, above and below the aromatic plane in addition to those corresponding to the two preferred enantiomer conformations. However, we have no direct evidence to support this.

Aromatic Frequencies. Three aromatic proton frequencies are discernible in all three solvents (Figure 2; Table 2). The NOESY cross-peaks to the meta protons of the aromatic ring (relative to the Ar$-$N) from the respective ortho substituents allows the meta and para protons to be observed selectively and is consistent with the asymmetric substitutions at the ortho positions. Conformational changes, for the aromatic ring and the attached ethyl and methyl groups, due to *S*- and *R*-chirality at the C-12 are not distinguishable in an achiral NMR experiments. However, conformers due to changes in the isopropyl methyl ether substituent could be observed in the NMR spectra.

Isopropyl Methyl Ether Frequencies. The three different conformations of the isopropyl methyl ether group (Figure 1a$-$c) can result in three different conformers for the H-12, H-13, $-O-CH_3$ (H-14), and H-15 protons that could be identified in the NMR spectra.

Two sets of doublets were observed for the H-15 protons in all three solvents (Table 2). Two-dimensional NOESY cross-peaks to H-15 protons were observed from the H-11 protons and the lower intensity H-10 protons. These data support the two preferred conformations c and b for a*S*,12*S*/a*R*,12*R* and a*R*,12*S*/a*S*,12*R*, respectively, since the spatial distance between the H-15 protons and the H-11 and H-10 protons in these conformations are conducive for NOE effects. This also verifies the two sets of triplets of H-10 protons, in Bz-d_6, as the two corresponding conformers of c and b.

Three sets of multiplets at different frequencies were observed for the H-13 protons in all three solvents (Table 2). NOE experiments conducted by irradiating

at the CH proton (H-12) confirmed that the two H-13 protons were split by the CH proton independently. The negative NOE effect at the high-frequency signals for H-13 protons, H-13c, and some of the H-13a frequencies indicates that these frequencies are due to the trans position proton of H-13 to H-12 proton. The multiple frequencies of H-13 were not resolved for the different conformations.

Binary Systems and the Chemical Environment. Metolachlor atropisomers were stable in all three solvents, but this was not the case in binary solvent systems. In pure Bz-d_6, the two H-10 frequencies appeared upfield to the H-15 frequencies. In a binary solvent mixture of CDCl$_3$:Bz-d_6, as the ratio increased, the H-10 frequencies coalesced (Figure 3). The frequency was not an average of the two frequencies; the signal was downfield to H-15, indicating deshielding. In pure CDCl$_3$, only one H-10 frequency was observed, and it was downfield from H-15.

The four H-8 frequencies in Bz-d_6 were upfield relative to the H-13 frequencies with the two least populated conformers appearing upfield to the other two, hence it was more shielded. Similar to the H-10 frequencies in a binary mixture of solvents, as the ratio of CDCl$_3$:Bz-d_6 increased, the four H-8 frequencies coalesced to two frequencies. These two H-8 frequencies were not averages of the four frequencies observed in Bz-d_6 and were downfield to H-13a, indicating deshielding.

At a 50:50 ratio, the H-10 and H-15 frequencies were indistinguishable, and the H-8 frequencies were downfield to H-13a (Figure 3). At a ratio of 65:35 (CDCl$_3$:Bz-d_6), only two conformers of H-8 were detected, and a single conformer for H-10 was obtained. This shows that the anisotropic interactions between the π-bonds of Bz-d_6 and metolachlor are evident when the Bz-d_6 solvent concentration is larger than $\sim$35%. In addition to the H-10 and H-8 frequencies, the H-11, H-14, and aromatic frequencies also showed a downfield shift from the frequencies in 100% Bz-*d6* when metolachlor was in 100% CDCl$_3$. The solvents did not influence all other isopropyl ethyl ether conformers.

Interconverting conformers can be observed as individual lines, broad lines, or a singlet depending on the chemical lifetime of the conformer. Figure 4 shows the influence of the DMSO$-$Bz binary solvent system on the proton spectra of metolachlor. Rapid exchange between the different conformers causes all the involved protons to become equivalent over time, and a single line is observed at the average chemical shift for the different conformers. The different conformers of metolachlor, observed in individual solvents (DMSO-d_6 and Bz-d_6) would broaden, coalesce, and become a single frequency at a Bz-d_6:DMSO-d_6 ratio of 40:60, indicating rapid exchange between the different conformers. Broad singlets were also observed for the protons H-10, H-9, H-15, H-12, and H-13 rather than multiplets due to coupling to adjacent protons. The two different conformers of the aliphatic protons H-15 and H-14 coalesced at a solvent ratio of 25:75 (Bz-d_6:DMSO-d_6) while the Ar$-$CH$_3$ protons (H-11) and CO$-$CH$_2$Cl (H-8) protons coalesced at a solvent ratio of 35:65 (Bz-d_6:DMSO-d_6). Single peaks were observed for the atropisomers a*S*,12*S* (a*R*,12*R*) and a*R*,12*S* (a*S*,12*R*) at the solvent ratio 40:60 (Bz-d_6:DMSO-d_6), indicating free exchange between the metolachlor atropisomers.

The rate of isomerization (k_c) for the exchange of unequal populations of the atropisomers a*S*,12*S* (a*R*,-
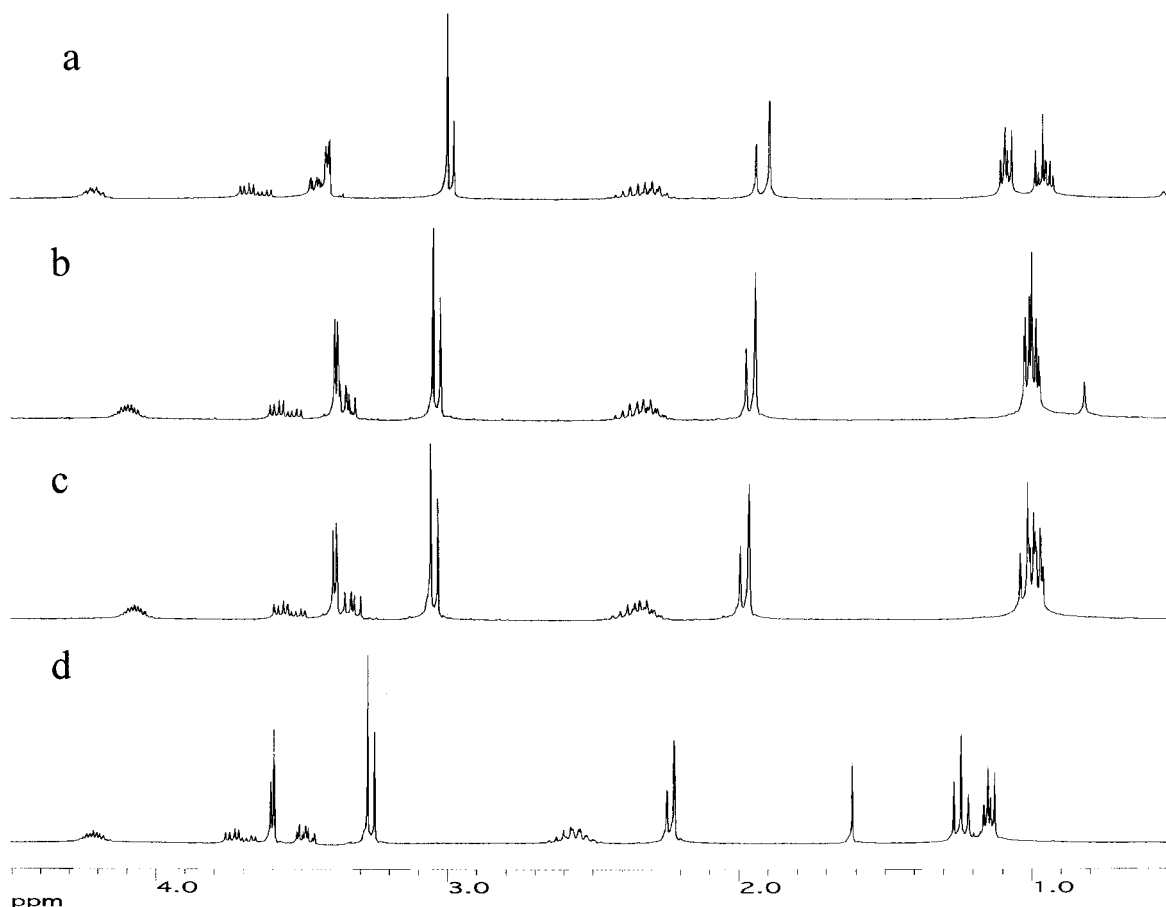
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Figure 3. Influence of a binary Bz-d_6 and $CDCl_3$ solvent system on the proton spectra of metolachlor conformers: (a) 100% Bz-d_6; (b) 50:50 ratio of Bz-d_6:$CDCl_3$; (c) 35:65 ratio of Bz-d_6:$CDCl_3$; (d) 100% $CDCl_3$.

12R) and aR,12S (aS,12R) was calculated using the following equations (Sandstrom, 1982):

$$p_a - p_b = \left(\frac{X^2 - 2}{3}\right)^{3/2}\frac{1}{X} \qquad (1)$$

where p_a is the population of atropisomer aS,12S (aR,-12R), p_b is the population of atropisomer aR,12S (aS,-12R), and

$$X = 2\pi\tau_c(\Delta\nu) \qquad (2)$$

$\Delta\nu$ is the difference in chemical shift of the exchanging atropisomers, and τ_c is the lifetime of the atropisomer aS,12S (aR,12R) in seconds was determined as 1.62×10^{-2} s. This lifetime is between 10 and 10^{-5} s and therefore is observed in the NMR time scale (McClure, 1996). The rate of isomerization of metolachlor atropisomers at 298 K was next calculated using

$$k_c = 1/\tau_c \qquad (3)$$

and was estimated to be 61.8 s^{-1}.

Transformation from slow exchange to rapid exchange between different configurations (racemarization) has been generally observed, in the past, with an increase in temperature (Moser et al., 1982; Buser and Muller, 1995). Metolachlor atropisomers have been shown to be stable at ambient temperatures, and interconversion between the isomers, in single solvents, has been postulated to be not possible even at 200 °C. However, this study confirms that, in specific chemical environments, free rotation/exchange between metolachlor atropisomers/conformers can exist at ambient temperatures (i.e., 298 K in this case) and that unequal populations of metolachlor atropisomers aS,12S (aR,12R) and aR,12S (aS,12R) can be made to be equal. Concomitantly, equal populations of aS and aR isomers result in chemical exchange between all the conformers that were otherwise conformationally distinct in pure solvents. Using a modified Eyring equation (Sandstrom, 1982):

$$\Delta G = 0.01914T[10.319 + \log(T/k_c)] \qquad (4)$$

where T is the temperature (K); the free energy of activation (ΔG) for the isomerization was determined as 62.8 kJ mol^{-1} in the Bz–DMSO (40:60) solvent system.

Influence of Water on Metolachlor Conformations. The fast exchange between the metolachlor atropisomers aS,12S (aR,12R) and aR,12S (aS,12R) observed in the Bz-d_6:DMSO-d_6 (40:60) binary system was slowed when a small quantity of D_2O was added (Figure 5). A slow exchange was also observed in a Bz-d_6:DMSO-d_6 (40:60) binary system when non-dry 99.6% Bz-d_6 was used (spectra not shown). However, the trace amounts of water associated with the DMSO-d_6 did not show any influence on the exchange rate of metolachlor atropisomers.

In inert solvents such as $CDCl_3$, molecules that possess labile protons such as O**H**, N**H**, and S**H** have broad line widths (and are not coupled to adjacent protons) due to intramolecular chemical exchange and hydrogen bonding. The addition of a polar solvent such as DMSO would slow intramolecular exchange due to
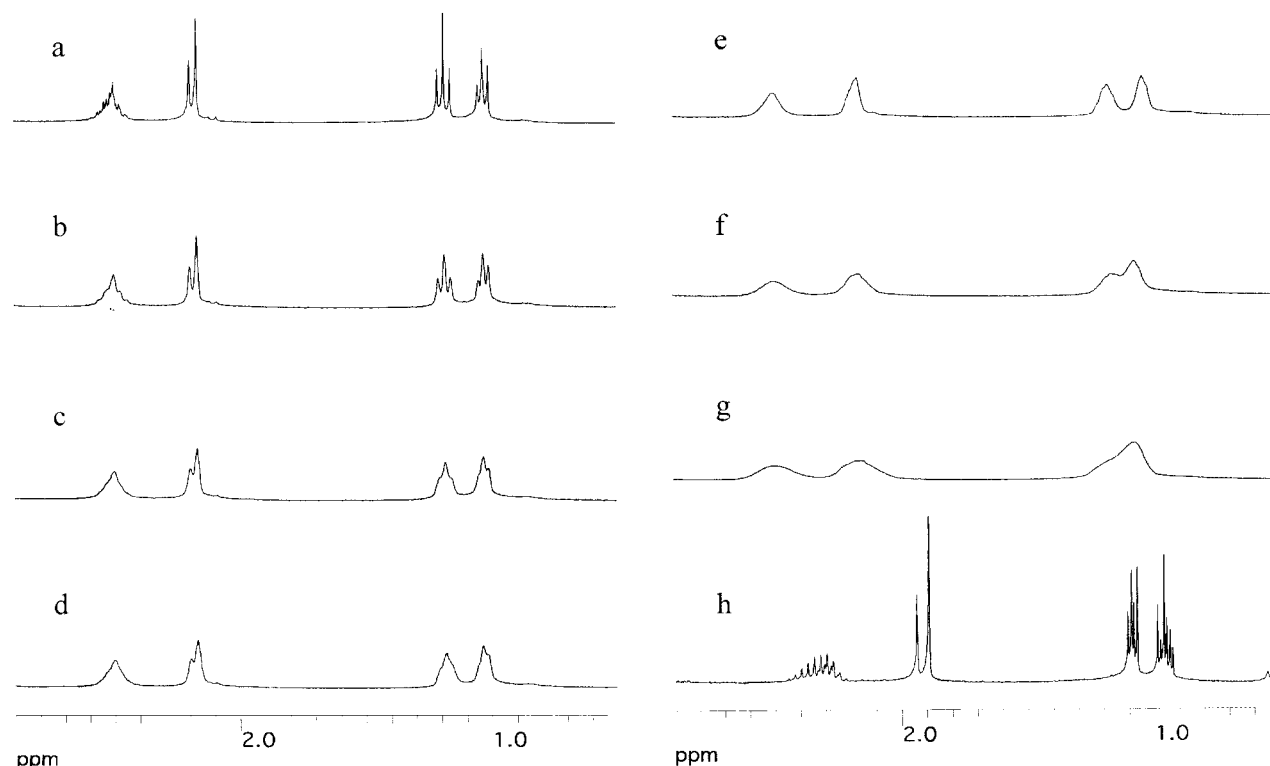
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Figure 4. Influence of a binary Bz:DMSO solvent system on the proton spectra of metolachlor conformers: (a) 100% DMSO-d_6; (b) 90:10 ratio of DMSO-d_6:Bz-d_6; (c) 75:25 ratio of DMSO-d_6:Bz-d_6; (d) 69:31 ratio of DMSO-d_6:Bz-d_6; (e) 60:40 ratio of DMSO-d_6:Bz-d_6; (f) 50:50 ratio of DMSO-d_6:Bz-d_6; (g) 40:60 ratio of DMSO-d_6:Bz-d_6; (h) 100% Bz-d_6.
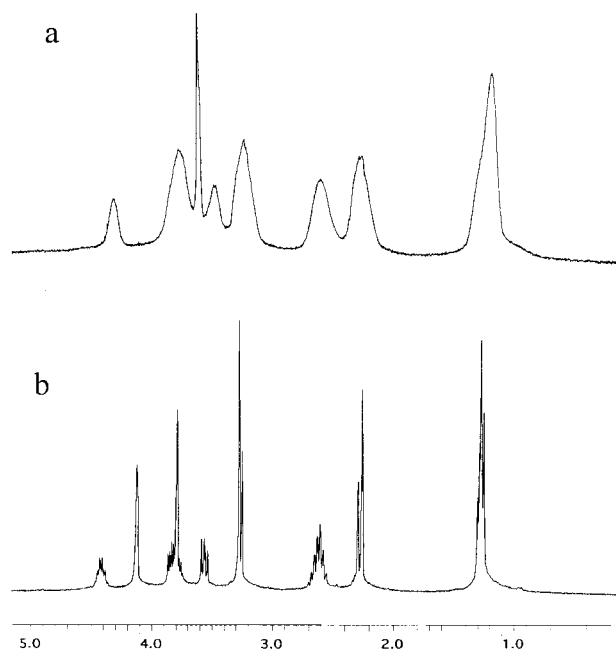

Figure 5. Influence of moisture on the proton spectra of metolachlor in a binary solvent system of Bz:DMSO: (a) 40:60 ratio of DMSO-d_6:Bz-d_6; (b) after the addition of two drops of D_2O to the mixture in panel a.

hydrogen bonding to the solvent. Yet, in the case of metolachlor no labile protons are identified in the molecule; hence, intramolecular chemical exchange cannot be expected. Furthermore, if labile protons were present when D_2O was added, the labile protons could be expected to vanish from the spectrum due to exchange with the deuterons. This behavior was not observed. Hence the line broadening observed in the Bz-d_6:DMSO-d_6 binary system is the result of rapid ex-

change between individual conformations of metolachlor atropisomers a*S*,12*S* (a*R*,12*R*) and a*R*,12*S* (a*S*,12*R*), which are slowed by water.

The resulting multiple individual conformations are thus stabilized by water molecules, due to possible hydrogen bonding to the C=O and −OCH$_3$ oxygen atoms. A molecular mechanics simulation conducted by solvating the metolachlor molecule suggested that metolachlor can form hydrogen bonds with water molecules at these sites. The conformational change or the change in molecular dynamics due to the addition of water results in the asymmetry of unequal a*S* and a*R* populations. This asymmetry then reestablishes the distinct unequal populations among conformations of the atropisomers a*S*,12*S* (a*R*,12*R*) and a*R*,12*S* (a*S*,12*R*) that we here assigned. This suggests that binding properties of metolachlor can be very different in symmetrical versus asymmetrical microenvironments or on symmetrical versus asymmetrical binding sites. Furthermore, the chemical environment also can influence the metabolism of metolachlor.

CONCLUSIONS

In this study, we were able to identify and resolve the solution conformations of metolachlor enantiomers a*S*,12*S*/a*R*,12*R* and a*R*,12*S*/a*S*,12*R*. Both aromatic and nonaromatic pure solvents stabilize unequal populations of distinct individual metolachlor conformations.

These individual conformations are destabilized in the binary solvent system Bz-d_6:DMSO-d_6, and rapid interconversion occurs between the metolachlor atropisomers a*S*,12*S* (a*R*,12*R*) and a*R*,12*S* (a*S*,12*R*), resulting in equal populations. The presence of a small amount of moisture in the Bz-d_6:DMSO-d_6 solvent system results in asymmetry of the metolachlor molecule due to possible hydrogen bonding and reestablishes the unequal,

individual conformations of metolachlor atropisomers. Hydrophobic binding sites, which preclude hydrogen bonding to the carbonyl site, can strongly effect the affinity of metolachlor. Similarly, aromatic pockets which effect aS and aR sites differently should also influence the binding of individual metolachlor isomers differently.

Our results suggest that in most natural environments (water is present) the different metolachlor conformations will be stabilized. Different chemical environments can stabilize certain conformations over the others, and this may influence the conformational ratios and the degree of degradation. If the less labile conformer is stabilized, then this would increase the persistence in the environment. Further experiments should be conducted to quantify rates of conformational changes in mixed solvent systems that closely resemble environmental samples where metolachlor is more likely be found. Environments that promote the fast dissipation of metolachlor and other organic pollutants will also set the basis for the successful remediation of conformationally stable compounds.

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